

Table II. Diagnostic codes and procedures

Insurer	Open Procedure	EVI	AKA/BKA	Admission For Gangrene
Private pay (PP) 21%	↓ 21%	↑ 62%	↓ 14%	↓ 13%
Medicare 68%	↓ 17%	↑ 101%	↓ 11%	↓ 5%
Medicaid 9%	↓ 24%	↑ 115%	↓ 14%	↓ 27%
Uninsured (UI) 1%	↑ 8%	↑ 38%	↑ 7%	↑ 35%
Significance	$P = .001$	$P = .03$	$P = .04$	$P < .001$

AKA/BKA; Above the knee amputation/below the knee amputation; EVI, endovascular intervention.

Hydrogen Sulfide Prevents Ischemia-Reperfusion Injury in Muscle even when Delivered in the Post-ischemic Period

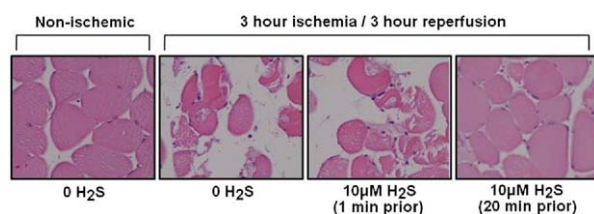
Henderson PW, Singh SP, Weinstein AL, Nagineni V, Spector JA

Objectives: Ischemia-reperfusion injury (IRI) is an unavoidable consequence of revascularization following acute vascular occlusion. Hydrogen sulfide (H_2S) is now recognized as an endogenous signaling molecule, and recent work from our lab has shown that H_2S mitigates IRI in muscle when delivered prior to the onset of ischemia. The purpose of this study was to determine whether H_2S confers a similar protective effect when delivered after an ischemic event has occurred.

Methods: Nine C57/BL6 mice underwent three hours of tourniquet-induced hind limb ischemia and were randomized to three groups: no H_2S , $10\mu M$ intravenous H_2S delivered one minute prior to reperfusion, and $10\mu M$ H_2S delivered 20 minutes prior to reperfusion. After three hours of reperfusion, the gastrocnemius and soleus muscles were harvested bilaterally. Sections were stained with hematoxylin and eosin (H&E) and subjected to a terminal deoxynucleotidyl transferase dUTP nick end labeling (TUNEL) assay. Values are reported as mean \pm SE, and student t tests were performed compared to non- H_2S -treated mice. Statistical significance was set at $P < .05$.

Results: Upon histologic examination, ischemic tissue from non- H_2S -treated mice had architectural changes including extracellular edema and intracellular vacuolization; tissue from mice treated with H_2S one minute prior to reperfusion was similar in appearance (Fig). Ischemic tissue from mice treated with H_2S 20 minutes prior to reperfusion showed no evidence of architectural changes. Non-ischemic tissue treated with H_2S was not different in appearance from non-ischemic, non-treated tissue. The percentage of apoptotic cells was significantly reduced by treating mice with H_2S 20 minutes prior to reperfusion, but not when H_2S was delivered one minute prior ($18.1\% \pm 11.0\%$ in non- H_2S -treated mice, $1.8\% \pm 1.4\%$ [$P = .026$] in mice treated with H_2S 20 minutes prior to reperfusion, and $16.0\% \pm 7.7\%$ [$P = .771$] in mice treated with H_2S one minute prior to reperfusion).

Conclusions: After an ischemic insult has occurred, H_2S protects against IRI when delivered 20 minutes, but not one minute, prior to reperfusion. This suggests the mechanism by which H_2S functions requires equilibration prior to reperfusion. The absence of injury in non-ischemic H_2S -treated tissue suggests that H_2S is non-toxic at the dose tested in this study ($10\mu M$). We conclude that H_2S may have clinical application as a component of therapeutic revascularization regimens.



Endothelial Cells with Eph B4 Heterozygous Knockdown: A Novel Model for Vein Graft Adaptation

Feigel A, Muto A, Fancher T, et al

Objectives: Vein graft adaptation (VGA) is characterized by vein wall thickening, a complex process requiring cell proliferation and migration. We have previously shown that VGA is also associated with loss of the venous determinant Eph-B4. To study molecular mechanisms that mediate VGA, we developed a novel in vitro model of VGA.

Methods: Aortic interposition grafts were examined using veins from wild type (WT) and Eph-B4 heterozygous knockout (Eph-B4-KO) mice.

Endothelial cells (EC) were isolated from both WT and Eph-B4-KO mice. Cell proliferation was directly counted. Quantitative polymerase chain reaction (qPCR) was used to evaluate basal mRNA expression. Western blot was used to evaluate protein levels. Cell migration was evaluated using a Boyden chamber.

Results: Vein grafts from Eph-B4-KO mice had 50% increased intimal thickness compared with vein grafts from WT mice ($P \leq .01$). Under basal conditions, Eph-B4-KO cells proliferated 41% more slowly than WT EC ($P = .029$), and 43% more slowly when stimulated with Ephrin-B2/Fc. Basal protein expression of Akt was 29% higher, and vascular endothelial growth factor (VEGF)-A was 28% lower in Eph-B4-KO cells compared with WT EC. Under direct stimulation, levels of phosphorylated AKT and phosphorylated ERK 1/2 were greater in Eph-B4-KO cells. qPCR confirmed 49% less VEGF-A mRNA expression in Eph-B4-KO cells. Eph-B4-KO cells had 60% reduced cell migration in response to Ephrin-B2/Fc ($P = .005$).

Conclusions: EC isolated from Eph-B4-KO mice serve as a novel in vitro model of VGA. This model suggests that reduced Eph-B4 expression during VGA may be mediated by upstream signals such as VEGF-A as well as downstream pathways such as Akt.

The Effect of Tortuosity on Five Different Cerebral Protection Devices in an Ex-vivo Model

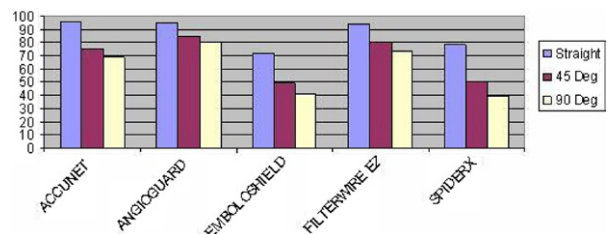
Shah AR, Lipsitz EC, Scher L, et al

Objectives: To compare the efficacy and ease of deployment for five currently available cerebral protection devices at varying degrees of tortuosity.

Methods: An ex-vivo study using procured canine carotid arteries was performed using an open flow model with three anatomical variants (non-tortuous, mildly tortuous, and severely tortuous) and two embolic particle sizes (250 and 425 microns). Cerebral protection devices (AccUNET [Guidant Corporation, Santa Clara, Calif], Angioguard [Cordis, Miami Lakes, Fla], Emboshield [Abbott Laboratories, Abbott Park, Ill], Filterwire EZ [Boston Scientific Corporation, Natick, Mass], and SpiderRX [cv3, Inc, Plymouth, Minn]) were placed under fluoroscopy at the point of maximal inflection in the case of tortuous vessels. A total of 1.5 mg (± 0.05 mg) of polyvinyl alcohol (PVA) particles of either size was injected into the carotid system proximal to each of five protection devices at each of the three degrees of tortuosity. Particles captured by the filters were weighed and compared with the weight of the particles collected in the effluent filter (not captured). The time taken to deploy each device at this position was recorded.

Results: The percent capture for each of the five filters at each degree of tortuosity using 425 micron particles is shown in the Fig. Similar results were obtained for the 250 micron particles. Filterwire EZ and SpiderX were deployed within the 26 to 27 second range. All others were deployed between 30 and 36 seconds.

Conclusions: No filter completely prevented embolization. Filterwire EZ, AccUNET, and Angioguard exhibited the highest capture rates in straight anatomy. Angioguard was the most efficacious filter in tortuous anatomy. Increasing tortuosity had a significant negative impact on all filters. Filterwire EZ and SpiderX had the best trackability in tortuous vessels using this model.



Mechanical Stimulation of HUVEC: Different Flow Patterns have Differential Effects on TF Expression

Nixon AM, Rochier A, Yamashita N, Sumpio BE

Objectives: Human umbilical vein endothelial cells (HUVECs) are known to respond differentially to various mechanical stimuli, and these different responses may be involved in atherogenesis. Here we investigated the differential effects of different flow patterns, such as laminar flow (LF) and orbital shear stress (OSS), on the expression of tissue factor (TF).

Methods: HUVECs were seeded on fibronectin coated glass slides and selectively in the periphery of gelatin coated culture dishes. When HUVECs on slides reached confluence, they were exposed to two, four, or six hours of LF (14 dynes/cm²). HUVECs in culture dishes were exposed to the same